

CLAIMS:

1. A method of producing isolated IgG1 subclass antibodies reactive to the surface of *Cryptosporidium* oocysts, the method comprising:
 - (a) pretreating *Cryptosporidium* oocysts with a reagent so as to remove the surface layer of the oocysts to form an oocyst antigen preparation;
 - (b) separating the oocysts from the oocyst antigen preparation so as to obtain a separated oocyst antigen preparation capable of eliciting a detectable IgG1 immune response in an animal to the surface of the oocyst;
 - (c) immunising an animal with the separated oocyst antigen preparation so as to elicit an IgG1 immune response in the animal; and
 - (d) obtaining from the animal IgG1 antibodies reactive to the surface of *Cryptosporidium* oocysts.
2. The method according to claim 1 wherein the reagent is a detergent.
3. The method according to claim 2 wherein the detergent is sodium dodecyl sulphate (SDS).
4. The method according to claim 3 wherein the pretreating is boiling the oocysts in the presence of SDS for a sufficient time to generate the oocyst antigen preparation.
5. The method according to claim 4 wherein (a) is boiling the oocysts for 1 hour in the presence of 0.5% (w/v) SDS.
6. The method according to claim 1 wherein the reagent is selected from the group consisting of urea, detergents including Triton X-100 and nonident, enzymes including chitinase, oxidising agents including sodium hypochlorite, sodium periodate, and ozone; and reducing agents including mercaptol ethanol and 1,1,1-trichloro- 2,2-bis[4-chlorophenyl]ethane.
7. The method according to any one of claims 1 to 6 wherein (c) further includes one or more adjuvants.
8. The method according to any one of claims 1 to 7 wherein the animal is a mouse.
9. A method of producing isolated IgG1 subclass antibodies reactive to the surface of *Cryptosporidium* oocysts, the method comprising:
 - (a) separating at least a portion of the *Cryptosporidium* oocyst wall from the internal sporozoites to form an oocyst-wall preparation;
 - (b) treating the separated oocyst-wall preparation so as to obtain an oocyst antigen preparation capable of eliciting a detectable IgG1 immune response in an animal to the surface of the oocyst;

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CLAIMS:

1. A method of producing isolated IgG1 subclass antibodies reactive to the surface of *Cryptosporidium* oocysts, the method comprising:
 - (a) pretreating *Cryptosporidium* oocysts with a reagent so as to remove the surface layer of the oocysts to form an oocyst antigen preparation;
 - 5 (b) separating the oocysts from the oocyst antigen preparation so as to obtain a separated oocyst antigen preparation capable of eliciting a detectable IgG1 immune response in an animal to the surface of the oocyst;
 - (c) immunising an animal with the separated oocyst antigen preparation so as to elicit an IgG1 immune response in the animal; and
 - 10 (d) obtaining from the animal IgG1 antibodies reactive to the surface of *Cryptosporidium* oocysts.
2. The method according to claim 1 wherein the reagent is a detergent.
3. The method according to claim 2 wherein the detergent is sodium dodecyl sulphate (SDS).
4. The method according to claim 3 wherein the pretreating ~~is~~ boiling the oocysts in the presence of SDS for a sufficient time to generate the oocyst antigen preparation. *comprises*
a *35/1*
a *C/1*
5. The method according to claim 4 wherein ~~(a) is boiling~~ the oocysts for 1 hour in the presence of 0.5% (w/v) SDS.
6. The method according to claim 1 wherein the reagent is selected from the group consisting of urea, detergents, including Triton X-100 and nonident, enzymes, including chitinase, oxidising agents including sodium hypochlorite, sodium periodate, and ozone; and reducing agents including mercaptol ethanol and 1,1,1-trichloro- 2,2-bis[4-chlorophenyl]ethane.
- 25 *a* *a*
a *a* 7. The method according to ~~any one of claims 1 to 6~~ *Claim 1* wherein ~~the preparation of step~~ further includes one or more adjuvants. *Claim 1*
8. The method according to ~~any one of claims 1 to 7~~ *Claim 1* wherein the animal is a mouse.
9. A method of producing isolated IgG1 subclass antibodies reactive to the surface of *Cryptosporidium* oocysts, the method comprising:
 - (a) separating at least a portion of the *Cryptosporidium* oocyst wall from the internal sporozoites to form an oocyst-wall preparation;
 - 35 (b) treating the separated oocyst-wall preparation so as to obtain an oocyst antigen preparation capable of eliciting a detectable IgG1 immune response in an animal to the surface of the oocyst;

(c) immunising an animal with the oocyst antigen preparation so as to elicit an IgG1 immune response in the animal; and
 (d) obtaining from the animal IgG1 antibodies reactive to the surface of *Cryptosporidium* oocysts.

5 10. The method according to claim 9 wherein the separation of the oocyst wall from the internal sporozoites ~~comprises inducing~~ is by causing the oocyst to excyst followed by immuno-separation of the oocyst wall components.

10 11. The method according to claim 9 wherein the separation of the oocyst wall from the internal sporozoite ~~comprises inducing~~ is by causing the oocyst to excyst followed by separation of the wall components by the group consisting of means selected from centrifugation, flow cytometry, density gradient separation, precipitation, immuno-labelling, ligand-binding, biotin-labelling ~~and~~ separation by avidin, and chromatographic separation.

15 12. The method according to claim 10 or 11 wherein causing the oocyst to excyst is by freeze-thawing or by physically breaking up by crushing, sonication, or grinding.

16 13. The method according to any one of claims 9 to 12 wherein the treating step (b) ~~comprises~~ is by physically breaking up the cell wall.

20 14. The method according to any one of claims 9 to 13 wherein (c) further includes one or more adjuvants.

15 15. The method according to any one of claims 9 to 15 wherein the animal is a mouse.

25 16. An isolated IgG1 antibody reactive to the surface of *Cryptosporidium* oocysts produced by the method according to any one of claims 1 to 8.

17. The antibody according to claim 16 ~~wherein the antibody is~~ being a monoclonal antibody.

18. An isolated IgG1 antibody reactive to the surface of *Cryptosporidium* oocysts produced by the method according to any one of claims 9 to 15.

19. The antibody according to claim 18 ~~wherein the antibody is~~ being a monoclonal antibody.

20 20. An isolated IgG1 antibody reactive to the surface of *Cryptosporidium* oocysts, the antibody ~~having~~ ^{wherein} has the oocyst binding and affinity characteristics of antibody CRY104.

21. The antibody according to claim 20 ~~being a monoclonal antibody~~ wherein

22. The antibody according to claim 21 being the IgG1 monoclonal antibody produced by ~~hybridoma~~ clone CRY104.

23. The hybridoma clone CRY104.

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